

### In the Specification

Please amend the paragraphs at page 8, lines 17-28, as follows:

Figure 1A. Stereo view of the overlay of engrailed HTH region ( $\alpha 2$ - $\alpha 3$ ; 1ENH) and one EF-Hand of parvalbumin (5PAL), to illustrate that the helical axes are colinear. The C-terminal  $\alpha 3$  is the homeodomain DNA-recognition helix. Engrailed is shown from the lower center, directly up and then to the right; parvalbumin is shown from the lower center to the lower left, then upwards and finally to the right; and the Ca(II) ion is shown as a solid circle having lines extended therefrom representing its ligands.

Figure 1B. A side by side stereo view of DNA (above) and a DNA (~~green~~) and metal binding (~~magenta~~) synthetic peptide (metal is shown as a sphere and synthetic peptide as a ribbon in blue) (below).

Figure 1C. Two views of the overlay of engrailed (1ENH) helix-turn-helix (HTH) region ( $\alpha 2$ - $\alpha 2$ ) and one EF-hand of calmodulin (1OSA; third Ca-site) thus illustrating that the helices occupy the same space. The C-terminal  $\alpha 3$  is the homeodomain recognition helix which binds in the DNA major groove.  $\alpha 1$ ,  $\alpha 2$  and  $\alpha 3$  of Engrailed (1ENH) are shown; and calmodulin (1OSA; third Ca-site) is shown binding a metal. The Ca(II) ion is shown as a solid sphere.

Please amend the paragraph at page 10, line 5, as follows:

Figure 8. Agarose gel electrophoresis showing the cleavage of pUC19 plasmid DNA by EuP3. Samples containing 1  $\mu$ g plasmid (about 60  $\mu$ M b.p.) in 10 mM Tris buffer, pH = 7.0, were incubated for 48 hours at 37°C. Reactions were quenched by the addition of 0.5 M EDTA to a final concentration of 0.1 M ~~40 mM~~, followed by addition of 100 mM KCl. The reaction mixtures were then treated with Amberlyst cation exchange resin prior to electrophoresis. Lane 1: pUC19 DNA control; lanes 2-5: DNA plus increasing concentrations of EuCl<sub>3</sub> (20, 30, 50 and 100  $\mu$ M); lanes 6-9: DNA plus increasing concentrations of free P3 (20, 30, 50 and 100  $\mu$ M); lanes 10-15: DNA plus increasing concentrations of 1:1 Eu:P3 (10, 20, 30, 40, 50 and 100  $\mu$ M). At the higher concentrations (lanes 13-15), significant affinity for DNA causes a decrease in total DNA intensity, likely due to aggregation causing insolubility.

Please amend the paragraph at page 48, line 21 as follows:

Metal binding and solution structure. The binding affinity of P3 for Eu(III) was characterized by isothermal titration microcalorimetry. The dissociation constant for EuP3 was found to be  $10 \pm 4 \mu\text{M}$ , from which the amount of bound and free Eu(III) in solution was calculated (Table 1). Though there is only one binding site per peptide, the binding behavior was not a simple two species process. EuP3 also dimerizes at higher concentrations  $K_{\text{dim}} \geq 80 \mu\text{M}$ . However, the second metal site in the dimer has low affinity ( $K_d > 1 \text{ mM}$ ), so free Eu(III), EuP3 monomer, and a singly occupied dimer ( $\text{EuP3}_2$ ), are the species present at concentrations below  $100 \mu\text{M}$ .

Please amend the paragraph beginning at line 1 of page 56 as follows:

Moreover, as shown in Figure 8, EuP3 catalyzes the cleavage of supercoiled, double-stranded DNA as well as model compounds. The conversion of supercoiled plasmid (type I) to open circular (type II), linear (type III), or smaller fragments was monitored by agarose gel electrophoresis. Because the synthetic peptides bind strongly to DNA, thus preventing the observation of products, the peptides were chelated prior to electrophoresis (Falke et al., 1994). At the point each reaction was quenched (0.1 M EDTA), a suspension of neutral, washed, cation resin (Amberlyst, 10-20  $\mu\text{L}$ ) was incubated with each sample for 30 minutes, spun down, and the supernatant loaded into wells. After 24 hours of reaction (incubated at  $37^\circ \text{C}$ ), the concentration-dependent formation of open circular plasmid was observed (Figure 8). Higher concentrations of EuP3 are less effective (slower), in keeping with the model BNPP system. Also of note is that 25  $\mu\text{M}$  EuP3 in the presence of 225  $\mu\text{M}$  excess metal (~~lane 3~~) had a similar effect to 25  $\mu\text{M}$  EuP3 alone, suggesting that peptide bound to DNA blocks indiscriminate Eu cleavage. Over a 10-300  $\mu\text{M}$  EuP3 gradient, nicking occurred from 10-150  $\mu\text{M}$ , with the greatest amount of cleavage at 30  $\mu\text{M}$  EuP3. EuP3 activity falls off with increasing concentration likely due to dimerization. Surprisingly, EuP4a activity does not.